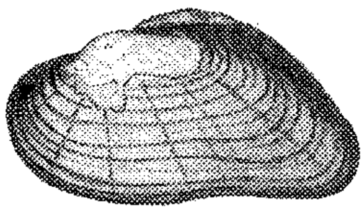
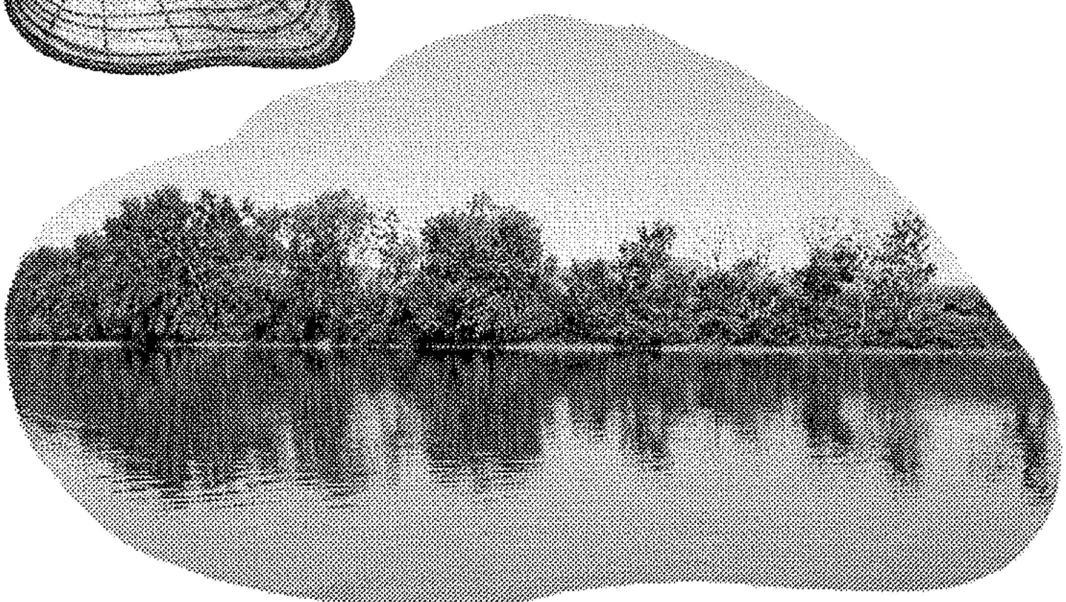
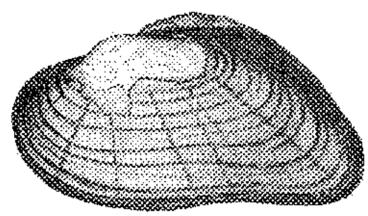
Nedeau et al 2003

QUANTITATIVE SURVEY OF THE DWARF WEDGEMUSSEL (*ALASMIDONTA HETERODON*) IN THE CONNECTICUT RIVER NEAR LUNENBURG, VERMONT







Ethan Nedeau

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QUANTITATIVE SURVEY OF THE DWARF WEDGEMUSSEL (*ALASMIDONTA HETERODON*) IN THE CONNECTICUT RIVER NEAR LUNENBURG, VERMONT

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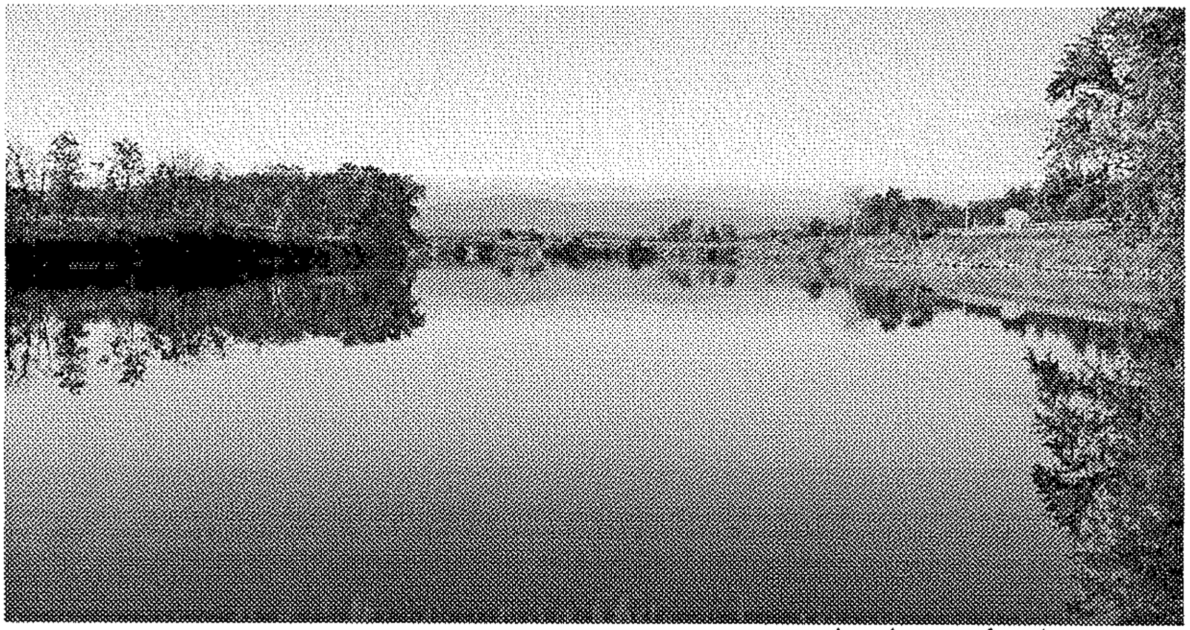
ABSTRACT

We performed a quantitative survey for dwarf wedgemussels (*Alasmidonta heterodon*) in the Connecticut River near Lunenburg, Vermont. The study design consisted of 4 sections, with 5-10 25m transects per section and 5 quadrats per transect, for a total of 145 quadrats, of which 49 were excavated to a depth of 10cm. We found 169 *A. heterodon*, for an overall average of 5.05/m^2. Densities ranged from 2.5/m^2 in Section 1 to 8.80/m^2 in Section 4. The population estimate for the survey area was approximately 6313 animals, and when projected to a quarter-mile section of river was nearly 182,000. Excavated quadrats yielded significantly more *A. heterodon* than unexcavated quadrats (7.28/m^2 vs. 3.88/m^2, $p<0.0001$), which supports prior assertions about the importance of excavation in quantitative surveys. Substrate exerted a strong influence on *A. heterodon* abundance, with animals over 5.5X more abundant in quadrats with >60% gravel than in quadrats with <20% gravel. Substrate may have also influenced the importance of excavation, because the difference between surface vs. buried mussel densities was greater in gravel substrates than in sand substrates. Observer bias affected the calculated densities and population estimates, and I recommend that researchers look more closely at this factor. Overall, this population is quite large and healthy—it is the largest yet documented in the Connecticut River and might be the largest remaining subpopulation of *A. heterodon* in the world. Long-term quantitative monitoring is recommended, though the study design could be modified to make the data more statistically valid and comparable with other studies.



Looking downriver from the survey area

INTRODUCTION

Long-term monitoring of freshwater mussels, with a goal of understanding population size or demography, requires quantitative and repeatable surveys. Qualitative surveys that document presence/absence and relative abundance provide very little insight into mussel populations, and will not provide future researchers with adequate baseline information to judge population trends. Throughout North America, efforts are underway to develop quantitative monitoring protocols so that populations can be compared across their entire range (Miller and Payne 1988, 1993, Downing and Downing 1992, Vaughn et al. 1997, Strayer et al. 1997, Smith et al. 2001a, 2001b).

The dwarf wedgemussel, *Alasmidonta heterodon* (hereafter referred to as DWM), is a federally endangered mussel that inhabits the Connecticut River and many of its tributaries. The upper Connecticut River between Charlestown and Lancaster, NH, support some of the largest and healthiest populations of DWM in the world. Past monitoring activity on the Connecticut River has focused on finding the extent of its range and documenting new populations. Since 1991 there have been several attempts to establish and maintain a long-term quantitative monitoring program for DWM in Windsor County (Fichtel 1991, Fichtel 1992, Fichtel 1993, Fichtel 1994, Gabriel and Fichtel 1995, O'Brien 2001). Despite earlier surveys, there is still not enough information about population size and demography with which to predict whether this species is declining in the Connecticut River.

The main objective of this project was to conduct a quantitative survey of a known population of DWM near Lunenburg, Vermont (Essex Co). A total of 536 DWM were relocated (87 were tagged) in 1997 prior to a bank stabilization project along Route 2, only 200 yards downstream of the current survey site (Gabriel et al. 1997). There have since been three follow-up surveys to determine survival of mussels, look for tagged mussels, and assess whether DWM were recolonizing the stabilization site (Gloria 1998, 1999, Gloria and Wicklow 2000). These studies have shown that DWM are quite abundant in this area—often exceeding 20 specimens per square meter—and that this reach may represent the best dwarf wedgemussel population remaining in the world. Thus, this DWM population provided an opportunity to establish a long-term quantitative monitoring program to document population trends.

This study was expected to provide the following types of information:

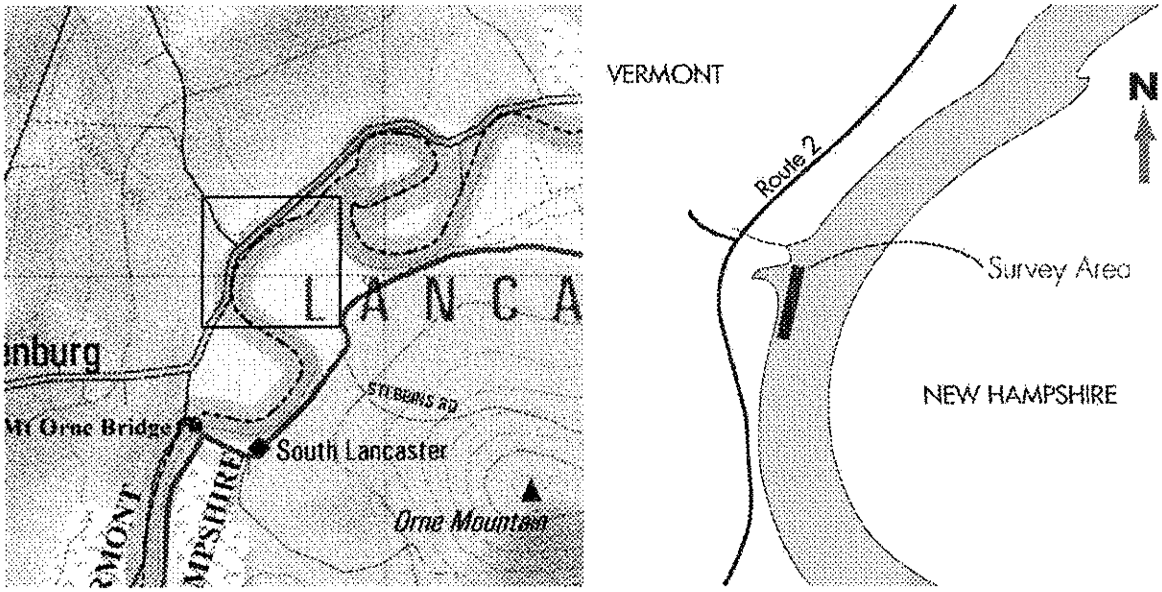


FIGURE 1. The Connecticut River betwen Lancaster and Lunenburg, and a close-up of the survey area.

- Abundance and density estimates for DWM
- Projected total abundance of DWM in the Connecticut River near the survey site
- Relationship between DWM distribution and substrate composition
- Size class distribution
- The importance of excavation on abundance and density estimates
- The significance of collector bias

METHODS

Study Site

The survey was conducted on the Connecticut River near Lunenburg, Vermont (Figure 1). The downstream end of the survey area was located approximately 200m upstream of the bank stabilization along Route 2, and extended upstream for about 120m. Surveys were confined to the west side of the river, within 30m of the shoreline. Surveys were conducted on August 22 and 23, 2002.

Study Design

The study design followed that of O'Brien (2001) to ensure data comparability. The basic design consisted of 4 sections comprised of 5-10 transects per section and 5 quadrats per transect (Figure 2, Appendix 1). Transects (25m) were established parallel to the stream bank, and 5 quadrats (0.25 m^2) were located at fixed positions along each transect (0m, 6.25m, 12.5m, 18.75m, 25m). Transects were positioned every 2m, and the number of transects in each section depended on habitat conditions (we usually began a new section once the substrate became 80-100% sand). Every third quadrat was excavated. Overall, there were 4 sections, 29 transects, and 145 quadrats, of which 49 were excavated (Table 1). The total area represented by the survey was 1250m^2, though the actual area surveyed (within the quadrats) was 36.25m^2

Survey Methods

Surveying was done with two divers and one person at the surface to transcribe data. Each diver worked side-by-side but independently in adjacent transects. Divers began at the downstream end of each transect and thoroughly searched each quadrat for mussels. All DWM were measured to the nearest millimeter using a ruler. In quadrats that were to be excavated, divers first counted and measured all animals at the surface, then excavated the entire plot to a depth of 10cm and looked for additional DWM in the sieved material. The number and length of buried animals was recorded separately from the surface data. In addition, divers estimated the relative abundance of substrate classes in each quadrat. : silt, sand, and gravel. Submerged macrophytes were sparse in the study area, wood was common but not included in the analysis,

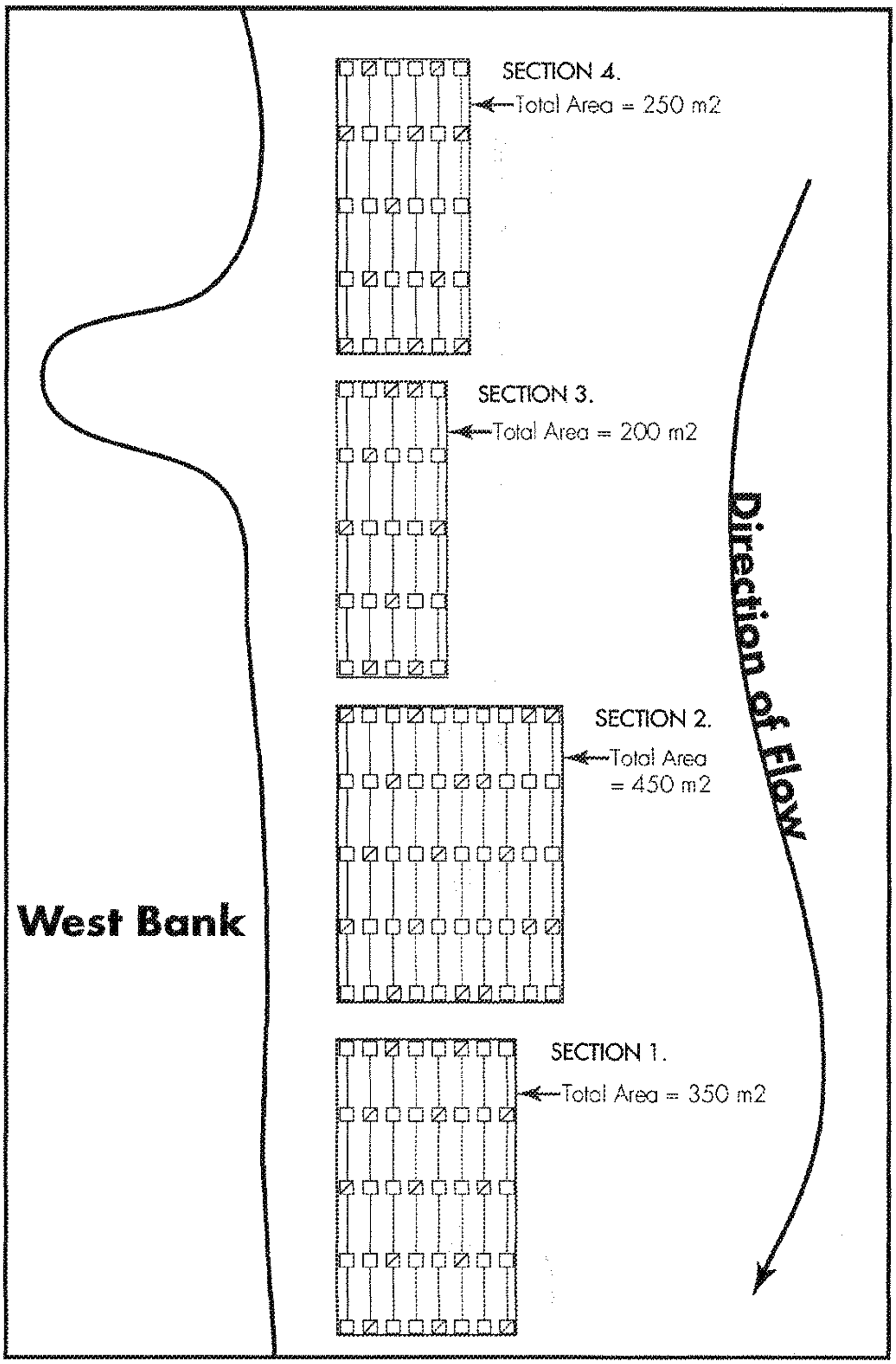


FIGURE 2. The approximate arrangement of sections, transects, and quadrats. Quadrats with a diagonal line were excavated. See Appendix 1 for actual numbers of DWM found in each quadrat.

and only silt, sand, and gravel were present in appreciable amounts.

Initially we intended to record the presence of all species, number of DWM, *Alasmidonta undulata*, and *Strophitus undulatus*, and lengths of DWM along each transect and within each quadrat. However, we found 47 DWM, 30 *A. undulata*, and 13 *S. undulatus* along the first transect alone. It took over an hour to complete the first transect. Since it was apparent that we would not be able to accomplish our study design objectives in a reasonable amount of time with this many animals to count and measure, we scaled back the survey by eliminating transect data and only counting and measuring DWM in the quadrats.

Analysis

All data were entered into a spreadsheet. Simple descriptive statistics were calculated to show density and abundance estimates, size class distribution, substrate relationships, collector bias, and importance of excavation. These results were displayed in tabular or graphic form. Some basic statistical tests were computed to examine the effect of collector, site, excavation, and substrate. Because of the nature of the data, the preferred test was the nonparametric Kruskal-Wallis analysis of variance, but in some instances a t-test or GLM analysis of variance was also computed. I used the SYSTAT 9.0 statistical package. The data was also run through USGS Mussel Estimation Software to compute population statistics. The data was modified to suit this program, since the study design was not the same as the design for which the software was written. All data is provided on the diskette that accompanies this report, and is either in MS Excel or WordPad.

RESULTS

Table 1 provides a summary of section, mussel, and habitat data. Raw data can be found in the Excel file that accompanies the report and is shown visually in Appendix 1. A total of 169 DWM were found, for an average of $5.05/m^2$. DWM were not evenly distributed across the four sections (Kruskal-Wallis test: $p<0.0001$, GLM test: $p<0.0001$). The densities of

TABLE 1. Summary of site, mussel, and habitat data.

DATA	SITE 1	SITE 2	SITE 3	SITE 4	TOTAL
Site Data					
# Transects	8	10	5	6	29
# Quadrats	40	50	25	30	145
# Excavated Quadrats	13	17	8	11	49
Total Area[a] (m^2)	350	450	200	250	1250
Mussel Data					
# Animals	25	45	33	66	169
# Animals/m^2	2.5	3.60	5.28	8.80	5.05[b]
Population Estimate	875	1620	1056	2200	6313[c]
Habitat Data					
Percent Gravel	19.7	41.4	48.0	59.2	41
Percent Sand	70.6	52.5	47.6	37.2	53
Percent Silt	9.7	5.1	4.4	3.6	6

a. This represents the total area encompassed within each site. NOT the area actually searched. The number of quadrats multiplied by 0.25 m^2 is the actual search area (=10, 12.5, 6.25, and 7.5m^2 for sites 1-4, respectively).

b. This value is computed by averaging sites 1-4, and thus gives equal weight to these sites despite varying sample sizes. A non-weighted value would be: (169/145) x 4 = 4.66 animals/m^2

c. See b2. This value is calculated based on the weighted average density of 5.05. A non-weighted total population estimate is 4.66 animals/m^2 x 1250 m^2 = 5825 animals.

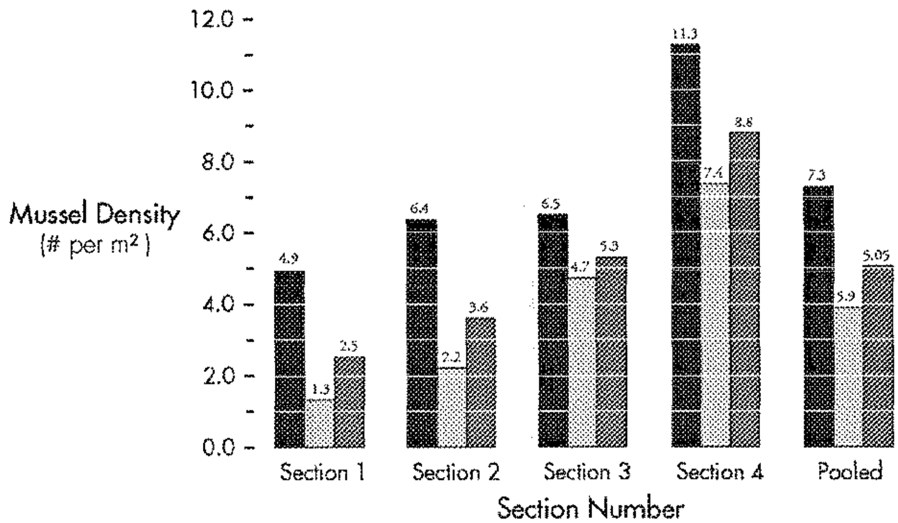


FIGURE 3. Mussel density at each section in excavated and unexcavated quadrats, and all quadrats combined.

DWM were lowest at section 1 (2.5/m^2) and highest at section 4 (8.8/m^2) (Figure 3: "Combined").

Excavated quadrats yielded significantly more DWM than unexcavated quadrats (Kruskal-Wallis test: $p<0.0001$) (Figure 3). This difference varied by section—excavated quadrats yielded 273%, 189%, 38%, and 53% more DWM than unexcavated quadrats in sections 1-4, respectively. For all sections combined, the density of DWM in excavated quadrats was 88% higher than in unexcavated quadrats, and thus the overall population estimate for the survey area (1250m^2) was also 88% higher for excavated plots (9100) alone than for unexcavated quadrats (4850) alone (Figure 4: "Combined").

The Mussel Estimation Software was used to compute population estimates for these data (Table 2), although these results are reported separately from the above data and should be interpreted with caution because the study design we used does not match the study design for which the program was written.

There was a collector bias (Table 3, Figure 4). Diver 1 found 104 animals (6.08/m^2) compared to 65 animals (3.96/m^2) for Diver 2. The Kruskal-Wallis test was not significant ($p=0.287$) and the t-test was marginally significant ($p=0.082$). Regardless, overall population estimates for the survey area based on Diver 1 data were 54% higher than for Diver 2. It is interesting to note that Diver 1 found 38% more mussels in excavated quadrats than Diver 2, but 82% more mussels in unexcavated quadrats. Since unexcavated quadrats require more careful and thorough visual searching, data suggests that the primary difference between the two divers was their ability to see and recognize DWM.

Substrate exerted a strong influence on results, particularly the relative abundance of sand versus gravel. The proportion of sand+silt was 80%, 58%, 52%, and 41% for sites sections 1-4, respectively (Table 1). In quadrats with 20% or less of gravel substrate, divers found an average of 1.56 DWM/m^2, whereas quadrats with greater than 60% gravel yielded nearly

TABLE 2. Results from the Mussel Estimation Software program.

STATISTIC	VALUE
Density	7.28/m^2
Standard Error	0.806
Coefficient of Variation	0.111
90% Confidence Limits	6.07-8.73/m^2
Abundance	9099
Standard Error	1007.60
90% Confidence Limits	7584-10917

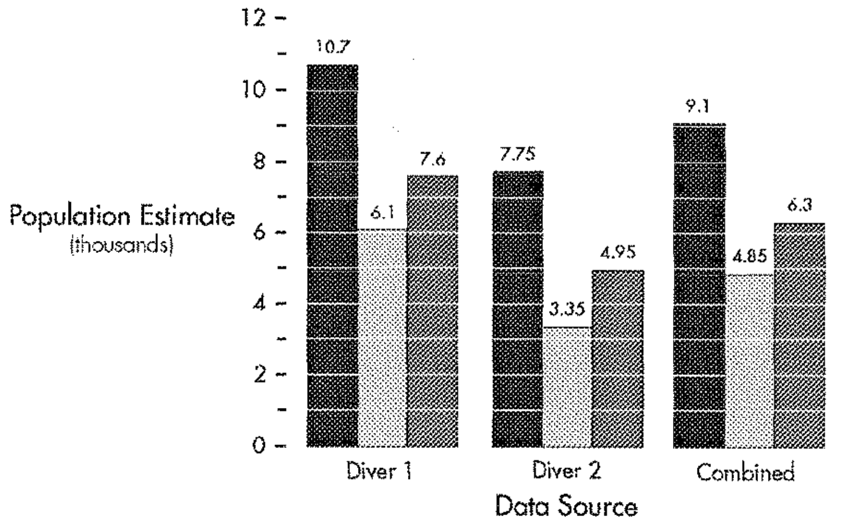


FIGURE 4. Population estimates for the survey area (1250 square meters) for each diver and both divers combined, for excavated plots, unexcavated plots, and all plots.

TABLE 3. Sampling effort, animals found, and population estimates for diver 1, diver 2, and the two divers combined (population estimates based on a total survey area of 1250 square meters).

OBSERVER	QUADRATS	ANIMALS	ANIMALS/M^2	ESTIMATE
Diver 1				
Excavated	24	49	8.56	10700
Unexcavated	51	55	4.88	6100
Combined	75	104	6.08	7600
Diver 2				
Excavated	25	38	6.20	7750
Unexcavated	45	27	2.68	3350
Combined	70	65	3.96	4950
Combined				
Excavated	49	87	7.28	9100
Unexcavated	96	82	3.88	4850
Combined	145	169	5.05	6313

TABLE 4. The effect of gravel abundance on DWM density and effectiveness of excavation.

	PERCENT GRAVEL			
DWM DENSITY*	0 - 20%	21 - 40%	41 - 60%	>60%
Average DWM/m²	1.56 (41)	5.54 (13)	4.76 (58)	8.86 (28)
Average DWM/m² in Excavated Quadrats	1.00 (12)	7.20 (5)	7.27 (22)	15.50 (8)
Average DWM/m² in Unexcavated Quadrats	1.80 (29)	4.52 (8)	3.24 (36)	6.20 (20)
Percent Difference in Excavated vs Unexcavated Quadrats	(-) 44.4	59.3	124.5	150.0

* Sample sizes in parenthesis. Totals are 5 shy of the total quadrats examined because substrate data was missing for 5 quadrats (2 that were excavated, 3 that were not).

8.9 DWM/m² (Table 4). Substrate may influence the importance of excavation, since our data shows that the proportion of gravel in a quadrat will affect the number of DWM at the surface versus in the sediment (Table 4). DWM may also be more visible at the surface in sand than in gravel. In quadrats with >60% gravel, density estimates in excavated quadrats were 150% higher than estimates for unexcavated quadrats (Table 4). In quadrats with 0-20% gravel, density estimates for excavated quadrats were 44% less than estimates for un-excavated quadrats.

The size-frequency histogram (Figure 5) indicates a healthy population, with 44% of the animals in the 30-34mm size range, and 85% of the animals in the 25-39mm size range. No juveniles less than 20mm were found, although this was likely due to a combination of sieve size and inability to confidently identify juveniles. Some juveniles were seen, but we were not comfortable distinguishing between small juvenile *A. heterodon*, *A. undulata* or *S. undulatus*. Surface animals were slightly longer than buried animals (Table 5), but this was not statistically significant.

The mussel assemblage at this site included *Elliptio complanata*, *A. undulata*, *S. undulatus*, and *Pyganodon cataracta*. *E. complanata* was the most common species, followed by DWM. However, our search image was biased toward DWM and we do not feel comfortable saying that it was more abundant than *A. undulata* or *S. undulatus*. We only found a handful of *P. cataracta*.

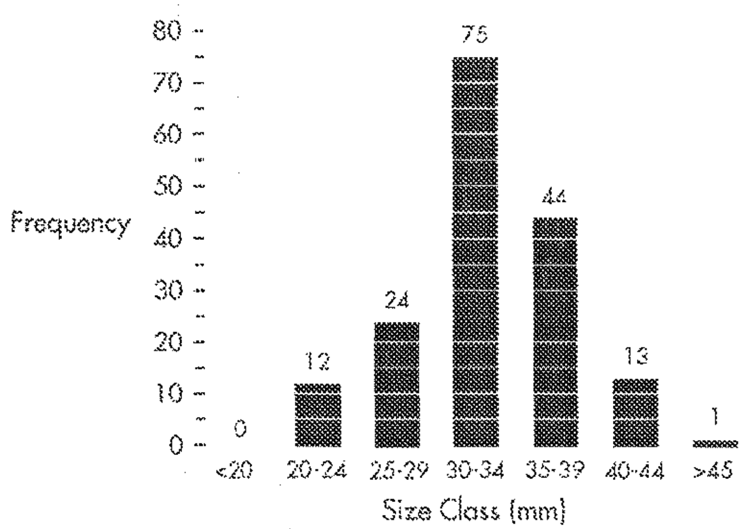


FIGURE 5. Size class distribution of DWM.

CONCLUSION

Densities of DWM in the study area are among the highest reported for this species throughout its range. A comparison of this site versus the other five sites that have been monitored by Vermont Fish and Wildlife (Cornish Covered Bridge North, Cornish Covered Bridge South, Horseback Ridge, Route 5 Cemetery, and Sumner Falls) shows that more DWM were found during this survey than were ever found at any of the other sites during 4-6 years of monitoring

TABLE 5. Length data.

LENGTH (MM)	SECTION				
	Sec1	Sec2	Sec3	Sec4	Pool
Mean Surface Animals	33.0	32.5	33.2	32.2	32.7
Mean Buried Animals	31.7	30.4	32.4	32.8	31.8
Mean Combined	32.5	31.7	33.2	32.3	32.4
Minimum Length	23.0	20.0	23.0	21.0	20.0
Maximum Length	48.0	41.0	41.0	40.0	48.0

(Table 6). However, neither the numbers nor densities are directly comparable because of different survey methods; earlier reports have used animals/person hour as an index of density. For comparison purposes, I calculated the DWM/person hour for the current survey based on number of air tanks used and an estimate of percent of time spent searching (much of our air was exhausted setting up and moving transect lines and relaying data to the recorder). This yielded an estimate of 14 DWM/person hour, which I still feel is very conservative because most other studies did not include excavation (Sumner Falls 2001 is the only other location where excavation was done), which also takes up a lot of time and air. Suffice it to say that this population is far larger than any population previously monitored on the Connecticut River.

Our density estimate of 5.05/m^2 and population estimate of 6313 DWM for the four sections represents a small area of the river (1250 m^2). The river is approximately 90 meters wide in this reach, and if we were to extend a line from bank to bank at the upper and lower end of the survey area, our survey area would only represent approximately 10-15% of the total area. Using our density estimate of 5.05 animals/m^2, we could extrapolate our results out to a larger area. We would estimate 181800 DWM for a quarter-mile section of river (400m length x 90m width = 36000 m^2 x 5.05 animals/m^2 = 181800 animals) and over 727200 for a mile of river. These projections, however, assume the spatial distribution of animals and substrates are similar throughout the entire reach (we know this assumption is likely not valid, but we did not take measurements to prove otherwise). A random sampling design over a larger section of river, paired with habitat data and measurements of river width, would be a better way to estimate population size.

Not surprisingly, gravel supported significantly more DWM than sand or silt. However, we still found quite a few DWM in sand and silt—more than we expected—and this should encourage us to rethink any preconception about habitat preference of DWM. The DWM-substrate relationship documented in this study could be compared to other studies to develop a model to screen projects for their expected impacts on mussels. However, our simple visual categorization of substrates is inadequate for building such a model, and more precise methods should be used for this purpose. Spatial mapping of substrates and mussel data using GIS could be a useful tool to visualize patterns.

Excavation was an important component of this survey. It is important that any quantitative survey include excavation of a subset of quadrats. Not much is known about the importance of excavation at different times of the year, though we know that many mussels move down into the substrate in the fall and resurface in the spring (Balfour and Smock 1995, Amyot and Downing 1997). The timing probably varies with water temperature, photoperiod, habitat, and the age, sex, or reproductive status of the animals. It would be worthwhile to conduct an excavation study in spring, summer, and fall to compare the importance of excavation—this might be important information for study design criteria when a survey must be conducted during a particular season. For example, if a construction project is to start in June and the survey must be conducted in May, it would be helpful to know approximately what percentage of the animals are expected to be buried or at the surface.

This study documents a fairly strong observer bias that is often not reported in the literature. The importance of observer bias cannot be overstated. Each

TABLE 6. Numbers of DWM and DWM/ person-hour reported from transect surveys at five long-term monitoring sites on the Connecticut River, 1991-2001. Current survey added for comparison purposes. See text for explanation of person-hours for the Lunenberg survey.
(Sources: Fichtel 1991, 1992, 1993, 1994, Gabriel and Fichtel 1995, O'Brien 2001)

SITE/YEAR	# DWM	DWM/person hour
Cornish Covered Bridge North		
1991	50	1.60
1992	0	0.00
1993	7	3.30
1994	4	2.80
1995	4	2.20
Average	13	1.98
Cornish Covered Bridge South		
1991	28	1.50
1993	22	7.20
1994	26	24.30
1995	10	3.40
2001	27	3.00
Average	22.6	7.88
Horseback Ridge		
1991	17	3.20
1992	26	5.30
1993	12	3.60
1994	18	8.60
1995	2	0.80
2001	28	4.70
Average	17.2	4.37
Lunenberg (This survey)		
2002	169	14.00
Route 5 Cemetary		
1991	7	3.30
1992	24	5.30
1994		10.80
1995	7	8.80
Average	12.7	7.05
Sumner Falls		
1991	7	1.40
1992	8	1.90
1993	9	5.30
1994	15	9.50
1995	14	7.40
2001	11	0.64
Average	10.7	4.36

observers effectiveness at searching for mussels is different, being determined by such factors as experience level, visual acuity, and patience. Excavation is perhaps a good means to overcome some observer bias, since finding mussels via excavation requires less "patience" and "finesse" than looking carefully for tiny mantle apertures. However, excavation requires more time and money, and it is not practical to excavate all quadrats. For particularly critical surveys where precision and accuracy are important, I would recommend that observers be subjected to a test to determine their relative skill at finding mussels, and the results then be used to adjust their estimates. I would like to know how other people have dealt with observer bias.

If this Lunenburg population is to become part of a long-term monitoring project, I would recommend that the state adopt the study design outlined in Smith et al. 2001a. Though there are countless ways to set up a statistically valid sampling design, they used a "double sampling design with 0.25 m^2 quadrats, systematically placed with multiple random starts, and excavation of a random subset of the quadrats". The USGS Mussel Estimation Software was written specifically for this design. Our study did not follow a double sampling design, nor did it include multiple random starts. The variance estimates and confidence intervals computed from our design were technically not correct. I corresponded with David Smith while analyzing the data (after the field work was finished) and we discussed his concerns about sampling design—basically, the extra work to implement David Smith's sampling design is in the planning stages, and it is very easy to implement in the field. Adopting the new design will make data more comparable with other studies conducted in eastern North America.

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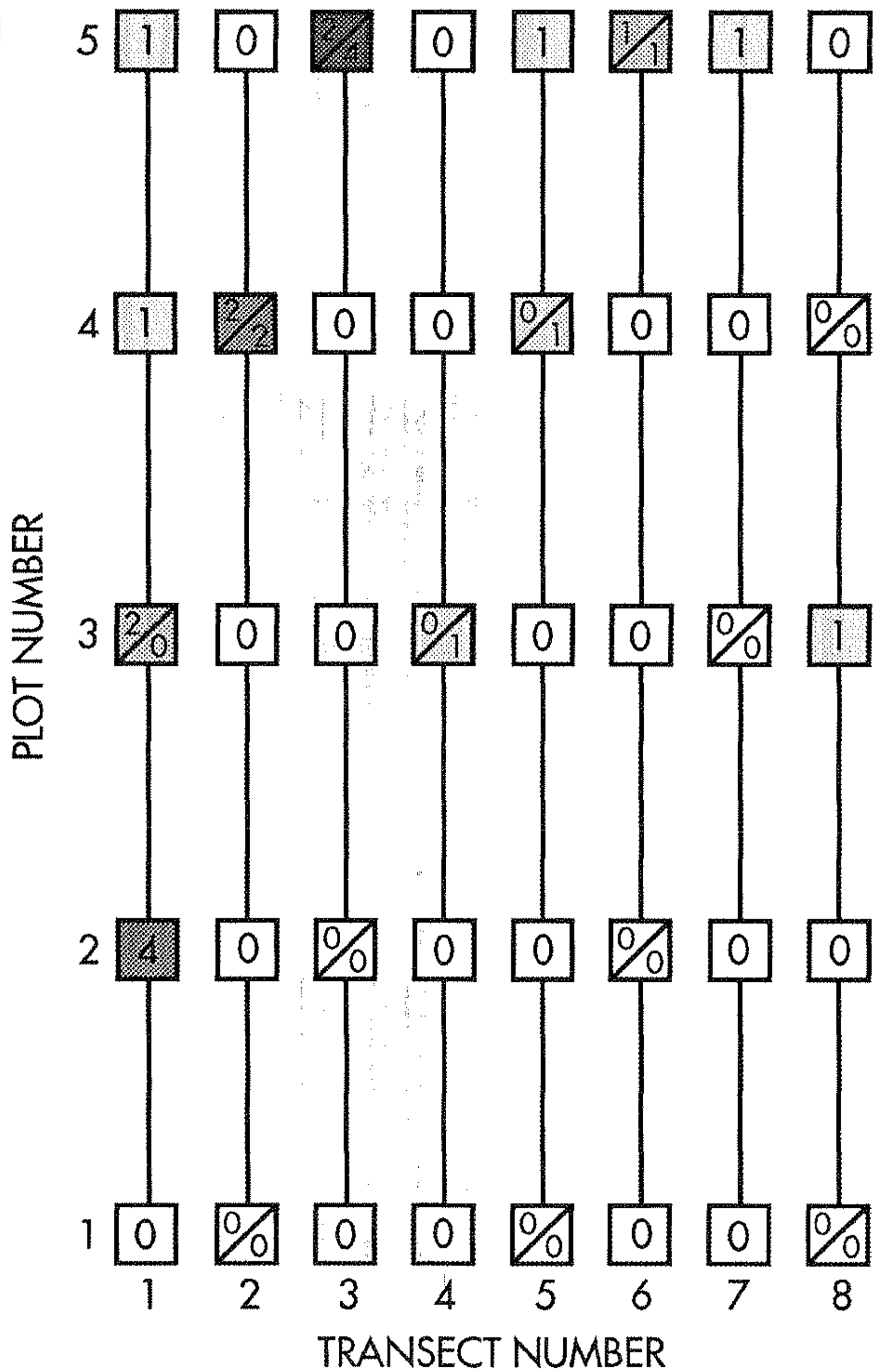
appendix one

SECTIONS, QUADRATS, AND MUSSEL DISTRIBUTION

These graphics show the arrangement of quadrats within each section, and gives the number of DWM found within each quadrat. Quadrats with a diagonal cross were excavated: the number in the lower corner indicates buried animals. Darker red indicates more mussels.

Section 1



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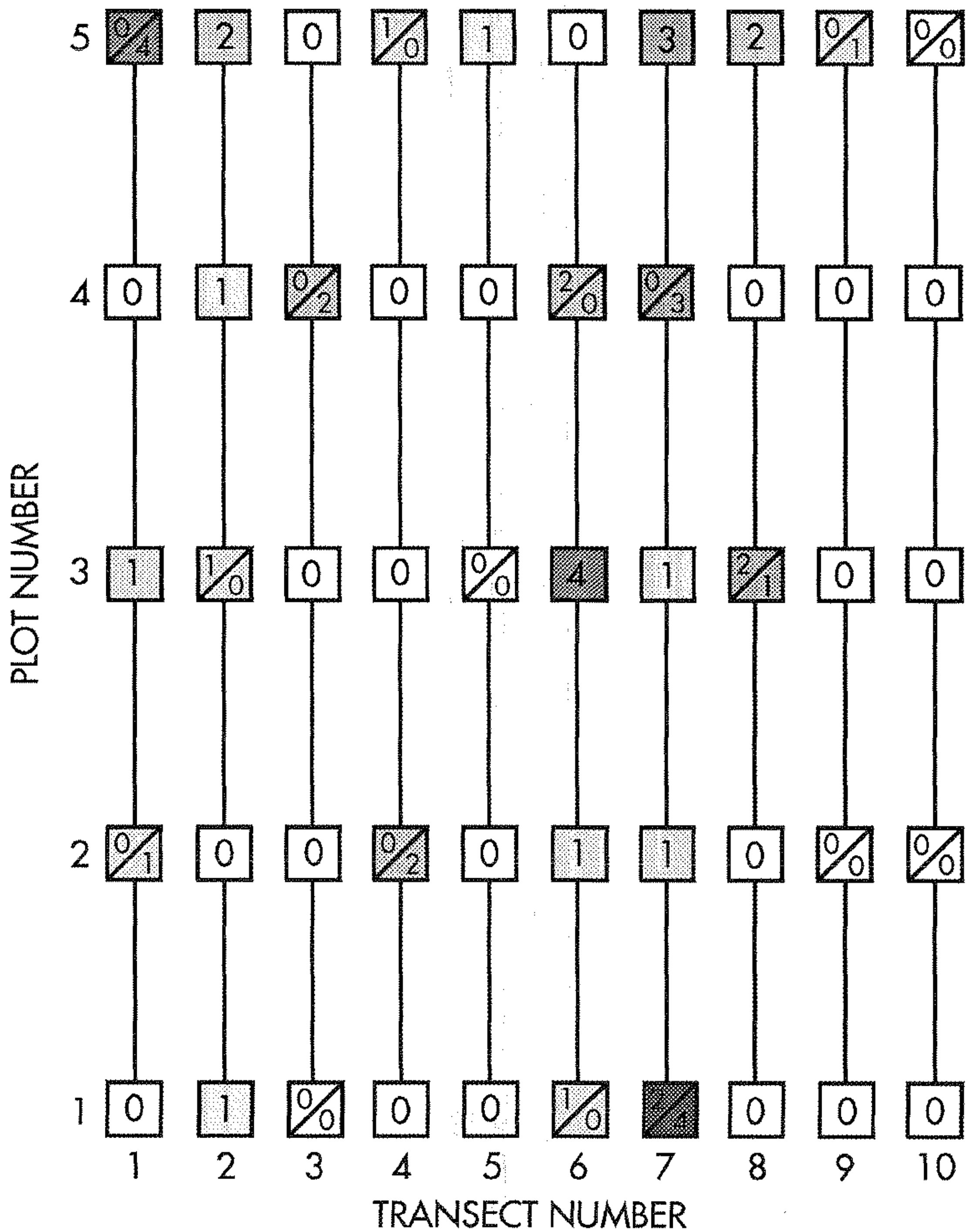

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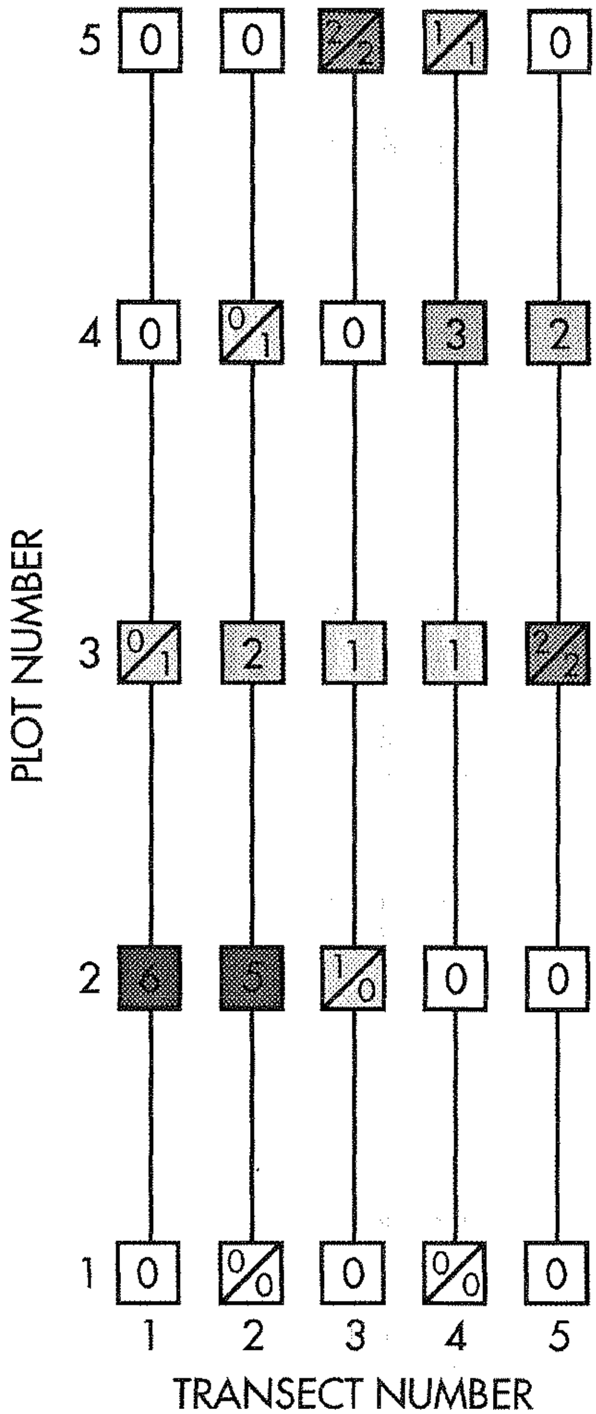

PLOT NUMBER
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3
2
1
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0
2/2
1/1
0
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0/1
0
3
2
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2
1
1
2/2
6
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0/0
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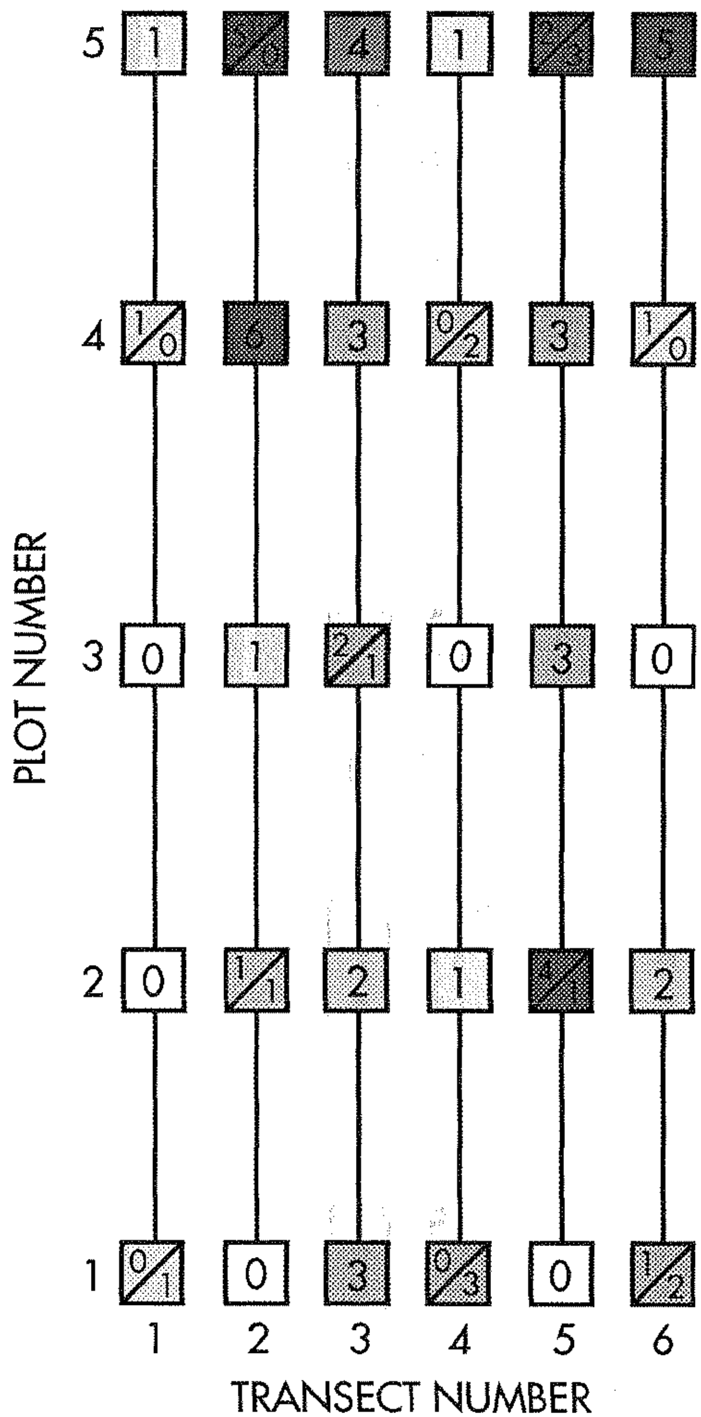
1
2
3
4
5
TRANSECT NUMBER

Section 4



Review of Survey Methods

Nedeau, E. J., M. Furgusop, and S. Werle. 2003. Quantitative survey of the dwarf wedgemussel *Alasmidonta heterodon*) in the Connecticut River near Lunenburg, Vermont.

Please note the following observations and comments regarding the methods employed in the above article.

- Surveys "were confined to the west side of the shoreline." Why was this? This needs to be explained to the reader.

- How and why were the survey sections selected? Was a stratification scheme used? Were the section positions randomly selected? Was the intent of the surveyors to generate estimates of known aggregations of mussels at the sites?

- Were quadrat positions within survey sections randomly selected? The descriptive phrase "fixed positions" was used in the methods, therefore it is logical to assume that random selection of quadrat positions was not employed. If the quadrat positions within survey sites were not randomly selected, then the density estimates may not be valid. Every quadrat within the survey section has to have an equal probability of selection.

- The authors state that "every third quadrat was excavated." Out of 4 survey sections, only 49 quadrats were excavated. Because of the low sample size number per survey section, the statistical accuracies of their estimates are probably quite low.

- The authors state that "because of the nature of the data" the non-parametric Kruskal-Wallis test was used. Then, they state that t-tests and the GLM procedure also were used. This needs to be explained in detail. When and why?

- What were the correlation coefficients and p values associated with the relationships between mussels found on the surface of quadrats and excavated from quadrats? Was there any relationship at all?

- Methods descriptions need to be more detailed to substanciate the validity of the extrapolation of findings to the proximate region of the Connecticut River. The reasons for site selections should be stated; and, the issue of random selection of quadrat positions within survey sites should be addressed.